
Deep Learning-Assisted Multiphoton Microscopy to Reduce Light Exposure and Expedite Imaging in Tissues With High and Low Light Sensitivity.

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Public Summary:

The purpose of this research was to use machine learning to analyze images with lower resolution and brightness. Thus, objects do not need to be exposed to high intensity laser for two-photon imaging. This will be less damaging and save time, especially for light-sensitive tissues such as retinal organoids.

Scientific Abstract:

Purpose: Two-photon excitation fluorescence (2PEF) reveals information about tissue function. Concerns for phototoxicity demand lower light exposure during imaging. Reducing excitation light reduces the quality of the image by limiting fluorescence emission. We applied deep learning (DL) super-resolution techniques to images acquired from low light exposure to yield high-resolution images of retinal and skin tissues. **Methods:** We analyzed two methods: a method based on U-Net and a patch-based regression method using paired images of skin (550) and retina (1200), each with low- and high-resolution paired images. The retina dataset was acquired at low and high laser powers from retinal organoids, and the skin dataset was obtained from averaging 7 to 15 frames or 70 frames. Mean squared error (MSE) and the structural similarity index measure (SSIM) were outcome measures for DL algorithm performance. **Results:** For the skin dataset, the patches method achieved a lower MSE (3.768) compared with U-Net (4.032) and a high SSIM (0.824) compared with U-Net (0.783). For the retinal dataset, the patches method achieved an average MSE of 27,611 compared with 146,855 for the U-Net method and an average SSIM of 0.636 compared with 0.607 for the U-Net method. The patches method was slower (303 seconds) than the U-Net method (<1 second). **Conclusions:** DL can reduce excitation light exposure in 2PEF imaging while preserving image quality metrics. **Translational Relevance:** DL methods will aid in translating 2PEF imaging from benchtop systems to in vivo imaging of light-sensitive tissues such as the retina.

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